The role of field experiments in the study of soil mediated effects of air pollution on forest trees

Felteksperimenter for å simulere langtidseffekter på skog av luftforurensninger

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Abstract

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Emissions of air pollutants can be looked upon as large scale experiments. Yet it is mainly in the case of local sources that dose-response relationsships can be studied with reasonable accuracy. Studies of the mechanisms behind the direct effects observed on plants are often best studied in short-term physiological experiments. Indirect effects, such as those connected with soil acidification, are more difficult to study because of their long-term character, often involving accumulative processes.

However, rigid criteria for causal relationships require that an observed disturbance can be duplicated by exposure to the suspected factor, and conversely that a malfunction can be remedied by removing the factor considered responsible. It is therefore necessery to consider if and how experiments on indirect effects of air pollution can be arranged.

The paper reviews results from mainly two factorial experiments in young forest of Scots pine, one with applications of sulphuric acid, fertilizers, and lime, the other with applications of nitrogen and PK fertilizer. The effects on the soils and on tree nutrition are briefly discussed, as well as the effects on tree growth.

The acid and nitrogen applications did increase the acidity of the mineral soil in both experiments, with a partial depletion of exchangeable calcium and magnesium stores. Magnesium concentration in the foliage was also decreased. Tree growth increased by 20 to 40 per cent after nitrogen application. Acid application without fertilization also increased tree growth, but much less, and for a limited period. With fertilization acid application decreased the response to fertilizer.

Even if not all of the a priori hypotheses of the experiments were confirmed, it is concluded that long-term ecosystem experiments on indirect effects of air pollutants are almost indispensible complements of short-term process studies. Such experiments may reveal otherwise unobserved interaction effects, both with time (reversal of initially positive effects), and between treatments (acid and fertilizer supply). In addition they provide possibilities to test the ecological realism of suggested causal relationships.

As the costs for ecosystem experiments are high, it is important that experiments laid out in the future build on already existing experience. This will insure that new experiments are placed on representative sites, and that plot sizes and numbers of replicates are large enough to permit the experiments to be kept under observation for at least some decades.

Key words: Air pollution, soil acidification, forest trees, field experiments.

Utdrag

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Publikasjonen resymerer resultater, spesielt fra svenske eksperimenter, der forsøksruter er tilført sure substanser, nitrogengjødsel og kalk. Virkningene på jord og trærnes næringssituasjon og vekst blir drøftet.

Den generelle konklusjon er at langtidsforsøk er det helt nødvendige verktøy i studier av effekter av deposisjon av luftforurensninger i terrestriske økosystemer. Dette til tross for høye omkostninger og tolkningsproblemer i dette kompliserte og heterogene økosystem, der f.eks. kalking først gjør seg gjeldende i dypere jordlag etter lang tid.

Det er spesielt viktig at fremtidige experimenter bygger på eksisterende erfaring, slik at forsøksarealene blir utvalgt på de mest følsomme boniteter, og at rutestørrelser og antall gjentak er stort nok til å følge forsøkene over årtier. Selv om mange utslag kan melde seg tidlig, f.eks. pH i de øvre jordlag, effekt på nitrogenomsetning og kationutvaskning, er trærnes vekst et resultat av kompliserte samspill og effekten av en bestemt behandling kan til og med endre retning over tid.

Nøkkelord: Luftforurensning, jordforsuring, skogstrær, feltforsøk.

Introduction

Ecologists have traditionally used two main methodologies for investigating the relations between organisms and environment: description of existing organism species and communities as well as their environment, and experiments changing one or more environmental factor on the other hand. Both these approaches – the comperative and the experimental – are indispensible, and there is an increasing understanding of the necessity of combining them. The purpose of the present paper is to discuss the role of experimental approaches in the study of indirect effects of air pollution on forest ecosystems.

Rigid criteria for establishing damaging effects of suspected adverse factors on plants were first formulated by Robert Koch in the 1870's. They are here quoted in the form given by COWLING (1988) in Table 1. Rule 1 deals with comparative evidence, and Rules 2 and 3 deal with experimental evidence; Rule 3 can be considered as a further elaboration of the two basic rules 1 and 2. Thus Koch more than a century ago required both comparative and experimental evidence to be convinced of a causal relationship.

Table 1. Koch's criteria for proof of causality in pathology, translated from German and in modern terminology. From COWLING1988.

Rule 1. The injury or dysfunction symptoms observed on individual organisms in the ecosystem must be associated consistently with the presence of the suspected causal factor(s).
Rule2. The same injury or dysfunction must be duplicated when healthy organisms are exposed to the suspected factor(s) under controlled conditions.
Rule 3. Natural variation in resistance and susceptibility of organisms observed in the ecosystem must be duplicated when clones of these same organisms are exposed to the suspected causal factor(s) under controlled conditions.

In the case of direct effects of air pollutants on plants, local emissions can be looked upon as large scale experiments. There are often reasonably good possibilities to study dose-response relationships as a function of the distance from the source, sometimes also as a function of time, if the time variations in the emissions are known. When further experiments are needed, they can be fairly simple in logical structure (though not necessarily in methodology), e.g. gas chamber experiments or transfers of plants to sites with different exposure.

Indirect effects of air pollutants on trees and other ecosystem components may in certain cases also be successfully studied along pollution gradients (FREEDMAN & HUTCHINSON) 1980, with further references). However, the complexity of the ecosystems, with many possible positive and negative feedbacks, makes the interpretation of such investigations difficult (TAMM 1976, 1989). We therefore have to look for additional approaches and consider their possibilites and drawbacks.

Ecosystem experiments

Indirect effects of air pollution on forest trees are such that affect the trees by changing their substrate or their biotic environment (competition, symbionts, conditions for pathogens, etc.). The relations between the trees and their biotic and abiotic environment concern ecosystem processes; an experiment changing one or more ecosystem process is then an ecosystem experiment. This does not mean that we should not carry out physiological process studies in the laboratory, but we also have to see whether results in the laboratory hold under field conditions, with all the various feedbacks in the ecosystem present. We have a sort of conflict here between the concept «controlled conditions» used by Robert Koch, and the complex ecosystem, where we do not have full control of all conditions. Does this conflict make

the experimental approach impossible, or can we find good or at least working solutions of the problem?

In table 2 we have listed some possible effects of air pollutants mediated by ecosystem components other than the trees but possibly affecting them.

Table 2. Ecosystem processes and characteristics of special interest in connection with deposition of acid and nitrogen.

FLOW CHART OF EVENTS	EXAMPLES OF CHANGES IN FLOWS OF ELEMENTS AND CHANGES IN PROCESSES
Deposition of air pollutants	Increased input of protons, mobile anions, bioelements, toxic substances, etc.
Changes in soil processes	Accumulation of deposited elements, changes in percolation pattern, changes in acid/base status, nutrient availability, soil biology, etc.
Changes in plant behaviour	Root damage, changes in nutrient uptake, carbon assimilation and allocation, stem production, etc.

We have to admit that the list of the various processes to the right above, is far from complete. However, if we for the moment consider it as covering the most crucial processes by which tree growth is related to soil acidification and nitrogen deposition, we can conclude that most of the changes listed can be studied more or less quantitatively by known methods. Nevertheless, it is clear that a very detailed list of processes to be studied makes our task a large ecosystem research program.

As we have limited resources for ecosystem experiments, we should start the experimental design from one or more welldefined and testable hypotheses on the effect of various air pollutants on forest ecosystems. A hypothesis for the behavior of a complex system is usually called a model. Models can be conceptual, described in words, or mathematical, described in formulas or diagrams. Working from a good a priori hypothesis gives important advantages, when it comes to statistical tests. But there are other advantages. We can also pinpoint important soil and plant processes which must be checked in order to see if the various steps in our model are changed in the expected way. The approach is laborious, but in the long run far more economic than the trial and error approach otherwise often used, e.g. in many old liming experiments. Also hypotheses other than those of the model can be tested

statistically in a properly planned experiment, given that the hypothesis to be tested is generated from independent evidence.

The following sections will give examples from the Swedish longterm experiments on soil acidification and increased nutrient supply, which have been designed from a priori hypotheses, though admittedly also with a component of trial and error. We believe that we have obtained results of interest not only where our hypotheses have been correct but also where they have been incorrect or insufficient.

The project «Accelerated acidification»

The experiments in this series began in 1969 as additional acitivity to the just-started project «Optimum nutrition experiments in forest stands» (see below). The background was the hypothesis presented by ODÉN (1968), that acid deposition acidified Swedish forest soils and depleted them of base cations, which in the long run should lead to a decrease in forest growth. We were sceptical of the hypothesis, knowing that the factor immediately limiting forest growth on mineral soil usually was nitrogen, but found it justifiable to include some acidification experiments in our program for study of forest nutrition (the optimum nutrition experiments). These first experiments – E42 Lisselbo and E57 Norrliden – certainly had an element of trial and error in them (if application of dilute sulphuric acid had no effect on the forest, we might as well forget Odén's hypothesis), but nevertheless started from the above-mentioned knowledge on nitrogen as a limiting factor and from Odén's base depletion hypothesis. If an effect were to be found, it was likely to be indirect, as pine and spruce are adapted during a long evolution to grow on strongly acid soils. It should then be possible to compensate adverse effects of acid by addition of nutrients. The experimental plan thus contained both acidity changes (by sulphuric acid or lime) and NPK fertilization in a factorial design (Table 3). The number of replicates were only two at Lisselbo, because of the limited number of plots available, but three at Norrliden.

The acid application, which was given as dilute, but still rather strong sulphuric acid, turned out to damage the lesser vegetation to an extent increasing from year to year. The acid applications terminated in 1976, after eigth years at Lisselbo and six years at Norrliden, while the fertilization continued (Table 3). The reason for ending the acidification was not only the vegetation damage. More important was that soil studies had shown differences in top soil acidity on both sites. It was then considered more important to study the after-effects, treatment differences in both soil and tree growth which could not be considered immediate artifacts of the annual applications of acid.

In 1976 we also had studied some processes and characteristics other than just pH on the treated plots (TAMM, POPOVIC, WIKLANDER 1977), with the somewhat unexpected result that, in short-term incubation experiments with mor samples, addition of acid or elementary sulphur increased nitrogen mobilisation (as ammonium ions). Acidification generally suppresses nitrification, but as neither Lisselbo nor Norrliden samples normally nitrified,

Table 3. Experiment E57 Norrliden. Total applications of acid and fertilizers 1971–1985. (From TAMM & POPOVIC 1989).

Treatment	Element kg/ha					
	S added as H_2SO_4	S added as PK fertilizers	Total amount 1971–85			
	1971–1976	1971–1985*	N	P	K	Ca
Control	–	–	–	–	–	–
Acid 1	98	–	–	–	–	–
Acid 2	196	–	–	–	–	–
Acid 3	294	–	–	–	–	–
Lime	–	–	–	–	–	2 000
N2P2K2	–	196	1 140	220	384	572
» + Ac 1	98	196	1 140	220	384	572
» +Ac 2	196	196	1 140	220	384	572
» +Ac 3	294	196	1 140	220	384	572
» + Lime	–	196	1 140	220	384	2 572

* SO_4–S content in PK-fertilizers, in 1971, 74, 77, 80, and 83.

this effect was of little importance. Incubation experiments with samples from Norrliden plots gave a somewhat different picture (HOLMEN et al 1976, POPOVIC 1984). There was a large variation between similarly treated plots, partly explainable by differences in C/N ratio. Addition of lime depressed total nitrogen mineralization (but increased nitrification on some fertilized plots), while fertilization appeared to have a moderate stimulating effect on total mineralization. At this date (1976) no clear effects of the acid treatments on tree growth had been observed, while fertilization had stimulated stem growth on both sites. Nor were there any clear effects of the acid on pine foliar element concentrations, while the NPK applications had increased the foliar nitrogen concentrations and also affected other element concentrations (TAMM & POPOVIC 1989). After 1976 the acid applications were discontinued, but not the fertilizer treatments. Annual sampling of current needles in autumn has continued, as well as tree growth measurements (diameter and height). Samples have also been taken for soil biology studies (BÅÅTH et al. 1980, 1984). In June, 1985, soil samples were taken with an auger from all plots in Experiment E 57 Norrliden down to 20 cm depth for chemical analysis of acid/base properties and exchangeable cations. Also increment cores have been collected from sample trees.

The results from the soil sampling in 1985 (table 4) confirm that the acid application really has acidified the soil, although the effect on the humus layer is not statistically significant, perhaps because the material in the humus layer is partly younger than eight years and thus not directly affected by the treatment. It is interesting to note that the NPK fertilization has acidified the mineral soil considerably more than the acid treatment. However, the

Table 4. Treatment effects on soil acidity in Experiment E 57 Norrliden. The acid effect is the arithmetic mean of all acid plots (approximately = Acid 2). The number of degrees of freedom (d.o.f.) for error in the ANOVA test is 16. Sampling in June 1985, nine years after the last acid application. pH is measured in water, TA (titratable acidity), exchangable calcium and magnesium in μeq per g D. W. The base saturation (V %) was calculated from the sum of exchangeable base cations and TA. Significance levels 5, 1, and O. I per cent marked with one, two or three asterisks respectively. (Data from TAMM & POPOVIC 1989).

Soil horizon and variable	D.o.F.	Difference in soil parameter				
		pH	TA	V %	CA_{exch}	Mg_{exch}
Humus						
+NPK/–NPK	1	+0.35***	–44	+5.7**	+18.7	–3.8**
+Acid/–Acid	3	–o.10	+60	–3.0	–0.7	+0.4
Acid x NPK	3	n.s.	n.s.	n.s.	n.s.	n.s
Mineral soil						
0 – 5 cm						
+NPK/–NPK	1	–0.11*	–4	+3.2*	+1.8**	–0.1
+Acid/–Acid	3	–0.11*	–1	–4.3**	–2.9***	–0.2*
Acid x NPK	3	n.s.	n.s.	n.s.	n.s.	n.s
5–10						
+NPK/–NPK	1	–0.42***	+11***	–2.6	–0.8	–0.2***
+Acid/–Acid	3	–0.19***	+1	–4.4*	–2.2**	–0.2*
Acid x NPK	3	n.s.	n.s.	n.s.	n.s.	n.s.
10–20 cm						
+NPK/–NPK	1	–0.50***	+5.3*	–3.6*	–0.8***	–0.1***
+Acid/–Acid	3	–0.21*	+4.6*	–5.2**	–0.9**	–0.2***
Acid x NPK	3	n.s.	n.s.	n.s.	n.s.	**

total amount of ammonium nitrate given before the sampling (Table 3) corresponds to 77 kmoles of nitrogen, while Acid 2 corresponds to a total amount of 6 kmoles of sulphur (12 kmoles of protons). It is also worth noting that the effect of NPK on pH in the humus layer is opposite to that in the mineral soil.

The long-term effects of the acid treatments on the needles were decreased magnesium concentrations, in agreement with the decrease in exchangeable magnesium in the soil. NPK application, on the other hand, affected the concentrations of most elements other than nitrogen in a negative direction. The PK addition was not enough to compensate for the «dilution effect» caused by nitrogen, and for the increased leaching of some elements.

The growth results are presented in Fig. 1 as relative stem volumes. There is a considerable variation in directly measured growth both within and between blocks, masking much of the treatment differences in direct comparisons. The data have therefore been adjusted by means of analysis of covariance fof difference in conditions at start as described by the covariate Bjørgung's index ($\sqrt{n} \times H^2$, BJØRGUNG 1968).

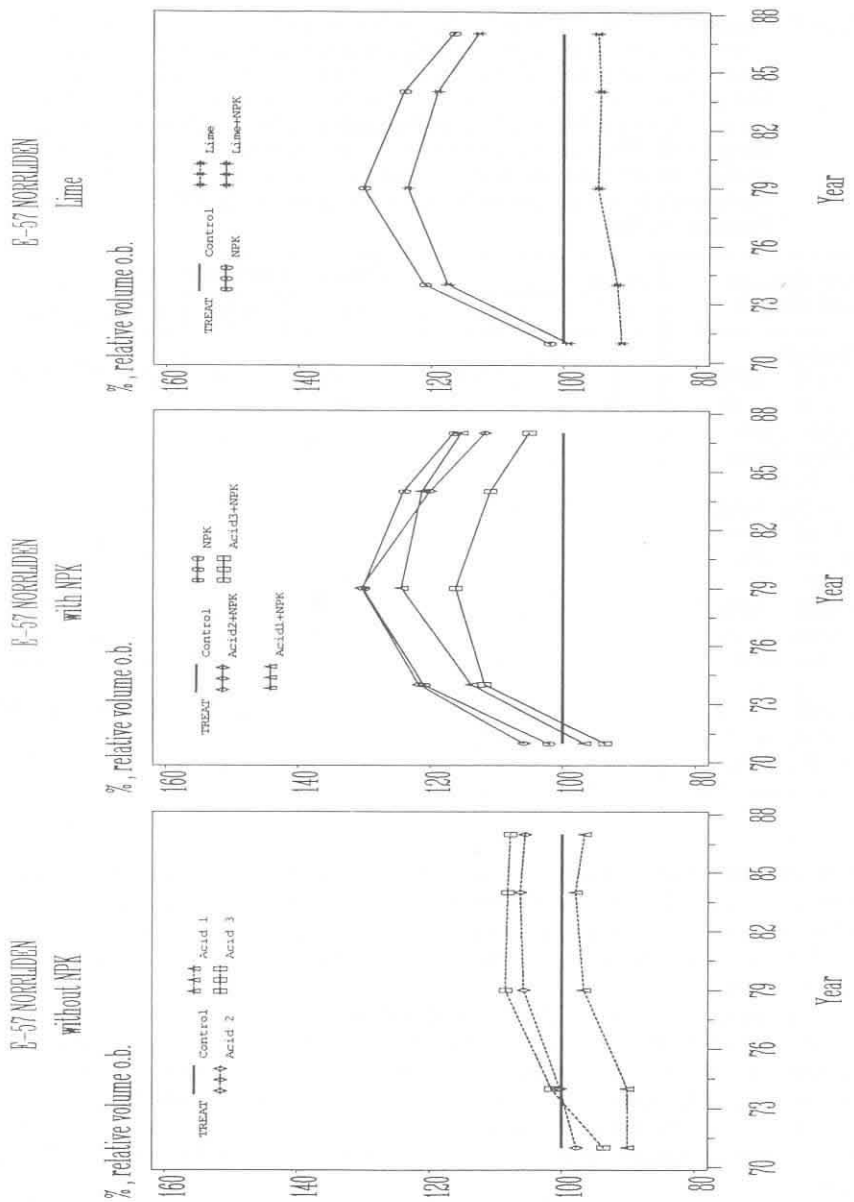

Fig. 1. Relative total stem production over bark in Experiment E57 Norrliden. Average of three control plots = 100. with standing volume 31 $m^3 \times ha^{-1}$ in 1971 and total production (standing volume +some self-thinning) 177 m^3 in 1987. Values adjusted by analysis of covariance for differences in Bjørgung's index, $\sqrt{n} \times H^2$ (n = number of trees per plot in 1970, H mean height in 1970). A. Treatments with supply of acid only. B. Treatments with acid and fertilizer. C. Treatments with lime, with and without fertilizer. From TAMM & POPOVIC (1989).

Apparently part of the between-treatment variation remains after the statistical adjustment in Fig. 1. Treatments showing positive or negative deviations in 1971 (before notable effects of the treatments) have a tendency to maintain their positions with respect to related treatments. Therefore the slopes of curves are more interesting than their levels. It can be seen that all NPK curves deviate positively from the controls, with a levelling-out after five to eight years. Acid without NPK also gives a rising trend in the beginning. The application of lime has resulted in curves paralles to those without lime (= no clear lime effect on growth). Together with NPK, higher levels of acid give a decrease in basal area and stem growth compared with NPK alone, most pronounced at the end of the measuring period.

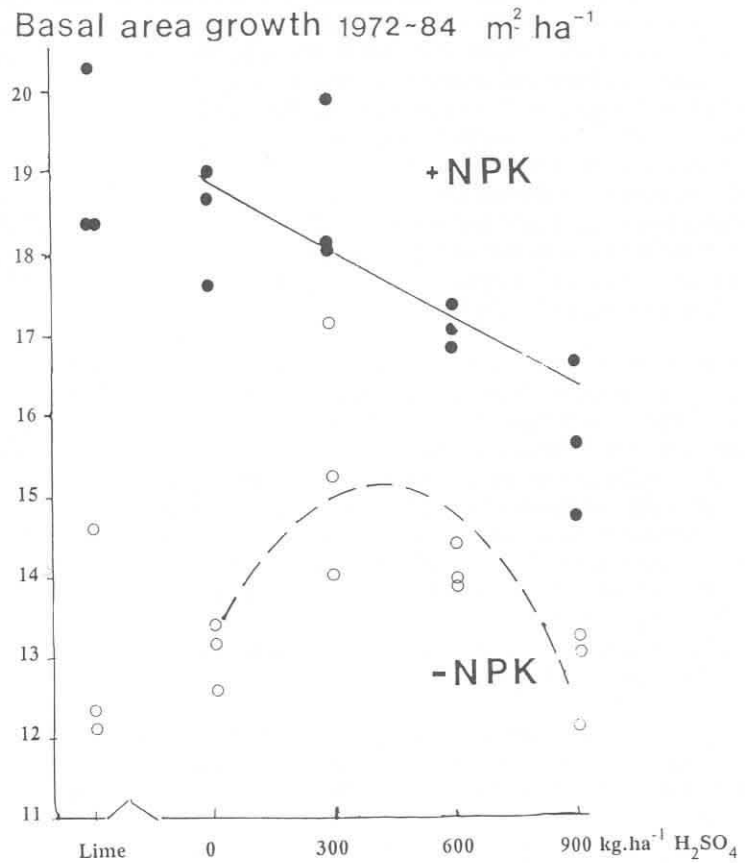

Fig. 2. Basal area increment 1972–1984 in Experiment E57 Norrliden, measured on increment cores. Individual plot values adjusted for differences in basal area growth 1966–1970 (before treatment) by analyses of covariance. Pretreatment basal area growth and treament (Acid 0-3) explains 64 per cent of the total variation in the dependent variable on both unfertilized plots and fertilized plots, and the variable Acid is statistically significant on fertilized plots, the variable $(Acid)^2$ on unfertilized ones. From TAMM & POPOVIC 1989.

Although relative values of standing volume provide an illustrative picture of the development, they are not easily tested statistically. We have earlier published basal area growth values for the period 1972–1979 (masured by calipering); a significant increase in basal area growth with increasing amount of acid on non-fertilized plots was found, as well as a decrease with acid on fertilized plots (TAMM & WIKLANDER 1980). New tests on the growth data 1972–1984 confirm the earlier conclusion with slight modifications. For stem volume increments, both the positive and the negative reaction appear similar to the basal area reaction in 1972–1979, but only the positive relationship is statistically significant (TAMM & POPOVIC 1989). In the case of the basal area increments, where the covariate (basal area growth 1966–1970) in the analysis of covariance provides an efficient adjustment for initial differences, both relationships (for growth versus acid, with and without fertilizers) are significant, but without fertilizers a curved regression line gives a much better fit than a straight line (Fig. 2). The slight discrepancy between Fig. 1 and Fig. 2 is thus that the latter shows a growth decline for treatment Acid 3, while in Fig. 1 Acid 3 still maintains a high growth-rate. As ring measurements are more accurate that calipering, and pretreatment growth, measured on the same cores, is a more effective adjustment variable than Bjørgung's index, the picture in Fig. 2. is probably the more reliable one.

Interpretation of experimental results is always difficult when there is large variation between plots, and a limited number of replicates. However, some conclusions can be drawn:

1. Application of a limited amount of sulphuric acid (6, 12 and 18 kmoles of charge per hectare) has acidified the mineral soil (and also, but more transiently, the humus layer), but only by 0.1 to 0.3 pH units, measured eight years after the last application. Exchangeable cations have also been leached, with the highest percentage loss for magnesium, followed by calcium. The immediate effect on tree growth has been stimulation, presumably due to descreased competition from microbes and higher plants. The growth stimulation levels out with time, and in the ring growth measurements there are signs of a reversal of the initially positive effect at the highest application.
2. Annual application of fertilizer has acidified the mineral soil even more than the acid application and increased growth considerably more than the acid.
3. The growth effect of fertilization is strong in the first years but then culminates. Later there is a tendency toward decreased differences beween fertilized and nonfertilized plots with time, in spite of the continued fertilization.
4. There is a strong negative interaction between acid and NPK. One possible explanation is that the combined soil acidification of both treatments has affected the trees negatively. Plots treated with high acid level and NPK have lost most of their original lesser vegetation (even if they are now invaded by *Deschampsia flexuosa, Chamaenerium angustifolium,* etc. where light conditions permit), so vegetational competition can hardly explain the development.

5. In this experiment there are no significant growth effects of liming, despite large changes in soil acid/base status.
6. As the pollution situation in large parts of Europa is dominated by deposition of both sulphur and nitrogen compounds, though usually not in the proportion given in the Norrliden experiment, interaction experiments between nitrogen and sulphur are more relevant for comparisons with the present situation than experiments with a single factor.
7. One of the basic assumptions at the start of our acidification experiments, viz. that NPK fertilization should compensate for adverse effects of acid deposition on nutrient status, could be rejected concerning the studied site with the tested form of acid application. However, a simple explanation of this result may be the strong acidifying effect of the NPK treatment.
8. On the other hand, another a priori hypothesis was confirmed, viz. ODÉN'S (1968) assumption that a limited amount of acid could further acidify a normal, already acid forest soil and partly deplete it of exchangeable base cations. The acid also appears to have changed the magnesium level in the foliage, and has affected tree growth. The effect was positive but probably transient, if given alone, and negative when combined with fertilizers. Both positive and negative growth effects have also been observed in Norwegian experiments (ABRAHAMSEN 1984, STUANES et. a. 1988).

Simulating nitrogen deposition in field plots

The original intention of our first optimum nitrogen experiment, laid out 1957 in a spruce plantation in southern Sweden (TAMM 1964, 1968), was to establish steady state conditions in the trees with respect to nitrogen nutrition, in order to get standard values for identification of suboptimum, optimum, and supraoptimum foliar concentrations. These cannot be derived from conventional fertilization experiments, where both nutrient concentrations and tree growth change rapidly with time. The results of the first trial showed that this type of fertilization experiment, establishing at least «quasisteadystate», could also be used to attack more basic questions about dynamic properties of forest ecosystems with respect to nutrition, growth mechanisms, allocation of nutrients within the tree, ultimate fate of added nutrients, etc.

With the increasing interest in environmental effects of forestry operations in the late 1960's it became urgent to start experiments where these basic questions could be studied in more detail. The early experiments had originally been planned for a five-year-period, with small plots and only two duplicates of each treatment. They were extended to a 13-year-period, but then had to be terminated because of plot size together with insect and drought damage. The clear-felling of the plots supplied interesting information on some of the environmental problems connected with large dosages of nitrogen fertilizers (TAMM & POPOVIC 1974)

The new experiments were not originally intended to simulate the atmospheric deposition of nitrogen, although already the creator of the Swedish, later European, network for precipitation chemistry measurements, Hans Egnér, was interested in the positive effects of nutrient input from the air. He was also aware of the partly anthropogenic origin of some of the rainwater nutrients. Yet ODÉN (1968) was the first one to establish clear time trends in rain acidity and sulphate concentration, as well as in nitrate and ammonium ions. He was disappointed not to find indications of positive growth trends in nitrogen-limited ecosystem related to the increase in nitrogen (ODÉN 1968)

Table 5. Fertilizer regimes in Experiment E55 Norrliden. The amounts are given as kg of element per hectare, broadcast on the ground in the beginning of the vegetation period. Nitrogen was given to different blocks either as urea or as ammonium nitrate. Phosphorus and potassium were given together as a commercial PK fertilizer. In 1980 the whole experiment received 2.5 kg boron (as borax) per hectare, since occasional symptoms of deficiency in boron had been observed on nitrogen fertilized plots. The experiments comprised six blocks, three with urea and three with ammonium nitrate, but two blocks (one with each form of nitrogen) were incomplete (lacking PK treated plots). (Data 1971–1984 from ARONSSON 1985).

Year	kg element per hectare to treatment:				
	N1	N2	N3	P	K
1971	60	120	180	40	75
1972	60	120	180		
1973	60	120	180		
1974	40	80	120	40	75
1975	40	80	120		
1976	40	80	120		
1977	30	60	90	40	78
1978	30	60	90		
1979	30	60	90		
1980	30	60	90	40	78
1981	30	60	90		
1982	30	60	90		
1983	30	60	90	40	78
1984	30	60	90		
1985	30	60	90		
1986	30	60	90	40	78
1987	30	60	90		

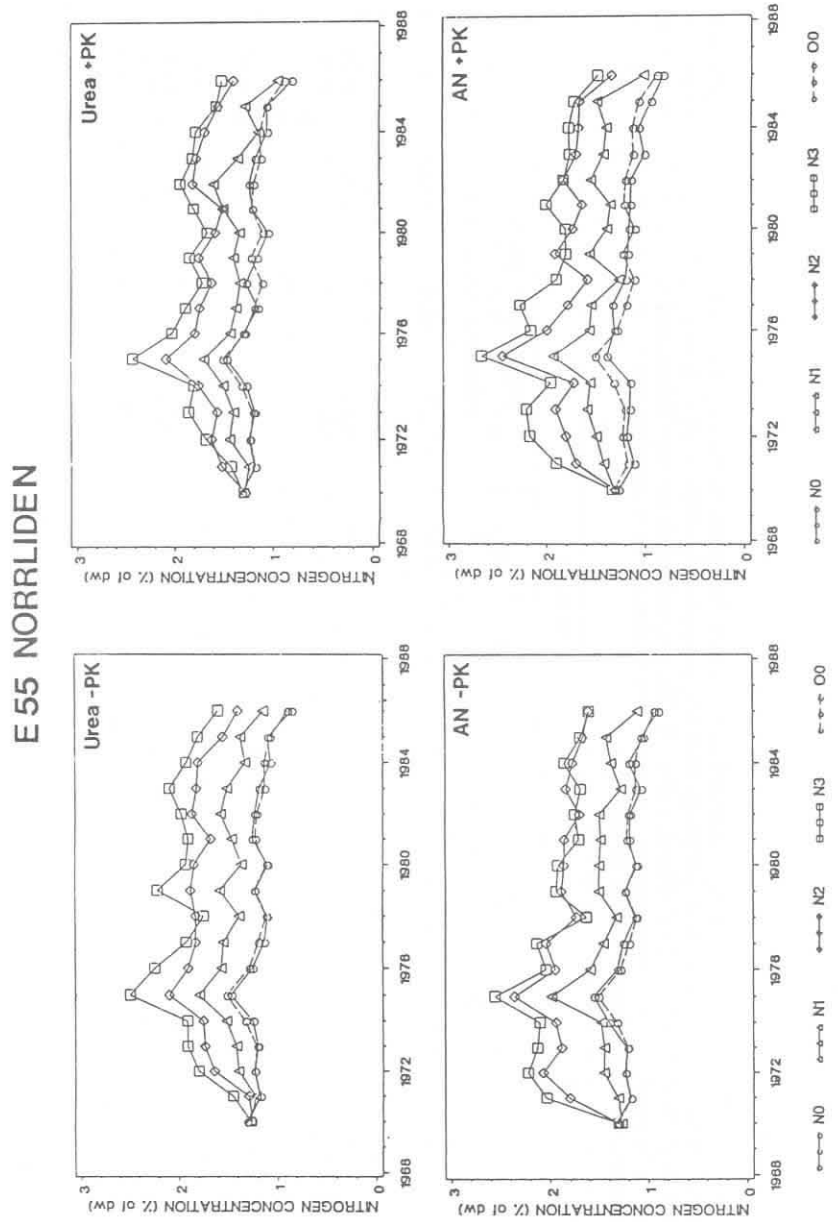

Fig. 3. Nitrogen concentrations in exposed current needles from Experiment E55 Norrliden. The symbols stand for different nitrogen regimes (see Table 5). Broken line (00), common for all four diagrams, stands for the mean of the six control plots, while NO in the left diagrams stands for the means of the three control plots in blocks fertilized with either urea (U) or ammonium nitrate (AN). In the right diagrams, NO stands for means of two PK plots in each diagram.

Table 6. Experiment E55 Norrliden. Soil acidity, measured as pH in water suspension, in June, 1988, in different soil horizons and with different fertilizer regimes. Data are given in colum 1 and 6 as aritmetic means of three replicate plots without any fertilizer (upper half Table), or for duplicate plots with PK only added (lower half Table), and in columns 2–5 and 7–9 as differences for the different nitrogen regimes from the NO treatment. LSD stands for the least significant difference at the five per cent level. (Data from TAMM et al. in prep.).

Soil layer	Treatment								
		Urea					Ammonium nitrate		
	N O pH	N 1	N 2	N 3	LSD	N O pH	N 1	N 2	N 3	LSD
		Difference from N O					Difference from N O			
Humus	4.48	+0.32	+0.33	+0.38	0.21	4.54	+0.27	+0.40	+0.40	0.29
Mineral soil										
0–5 cm	4.49	–0.04	–0.12	–0.22	0.14	4.68	–0.23	–0.29	–0.34	0.26
5–10 cm	5.08	–0.08	–0.58	–0.88	0.17	5.18	–0.25	–0.89	–1.07	0.21
10–20 cm	5.63	–0.03	–0.87	–1.25	0.32	5.60	–0.01	–1.03	–1.22	0.34
			with P2K2					with P2K2		
Humus	4.86	–0.06	–0.07	–0.08	0.25	4.78	–0.02	+0.06	+0.12	0.19
Mineral soil										
0–5 cm	4.49	–0.15	–0.18	–0.23	0.29	4.64	–0.22	–0.28	–0.35	0.17
5–10 cm	4.93	–0.25	–0.37	–0.62	0.34	5.04	–0.34	–0.75	–0.84	0.32
10–20 cm	5.26	–0.24	–0.40	–0.76	0.33	5,43	–0.35	–0.79	–1.04	0.42

In the series of field experiements within the project «Swedish Optimum Nutrient Experiments in Forest Stands», started in 1967 (TAMM 1985), we soon found that we could decrease the initially high annual additions of nitrogen and still keep the needle nutrient concentrations well above those of the control plots (Table 5, Fig. 3). From 1977 onwards the lowest annual nitrogen addition (N1) has been 30 kg nitrogen per hectare and year, a figure now often exceeded in areas of middle Europe with intensive agriculture and animal husbandry. Deposition figures from southernmost Sweden are approaching this value, so one of the present objectives of the project is to look for effects on the forest ecosystem of prolonged deposition of nitrogen.

As in the acidification experiment, we got a pronounced acidification of the mineral soil after fertilization (Table 6), and as Experiment E 55 contains treatments with both nitrogen and PK we can conclude that most of the acidification is due to nitrogen, not to PK. Both fertilizers appear to have increased the pH of the mor layer, while all nitrogen treatments have lowered the pH in the mineral soil , in the case of treatments N2 and N3 almost always statistically significantly. Remarkably enough there is not much difference between urea and ammonium nitrate; urea acidification must depend on nitrification, while acidification with ammonium nitrate can be explained by the mobile anion concept (SEIP 1980), nitrate ions percolating the soil, accompanied by protons from deposition or from the mor laver, and removing basic cations from the mineral soil. Increased forest growth has also been suggested as a cause of soil acidification, but there is no statistically significant difference in stem growth over the whole period between the treatments N1 and N2 (Fig. 4) ; treatments N3, with less stem growth, is most acidified. The limited acidification with treatment N1 Urea fits well with the observation by POPOVIC (1977) that N1 treatments did not nitrify in incubation experiments in 1974, and with results from HÖGLUND et al. (1986) who found almost as low levels of nitrate reductase in leaves of *Deschampsia flexuosa* from N1 Urea plots as in those from controls. All other treatments showed higher concentrations of this enzyme, indicating that the grass had access to soil nitrate, either from fertilizer (ammonium nitrate) or from nitrification (both types of fertilizers, but probably mostly on urea plots).

The additions of nitrogen fertilizer in higher rates have completely changed the lesser vegetation. *Deschampsia flexuosa* and *Chamaenerium angustifolium* dominate the field layer instead of dwarf shrubs, and also many of the original feather mosses and lichens disappear. Where the canopy is very dense, patches free of vegetation occur. A detailed investigation has recently been done, but all results are not yet available (DIRKSE & VAN DOBBEN , 1989).

The growth results, as those from Expt. E 57, are not entirely clear, because of the large between-plot variation. A good comparison between blocks fertilized with either ammonium nitrate or urea is difficult, as there seems to be some site differences between the blocks. While all plots were randomized within blocks, there is only one degree of freedom for the comparison between sources of nitrogen. However, we compared the relative growth data from the field measurements (fig. 4) with ring measurements from felled

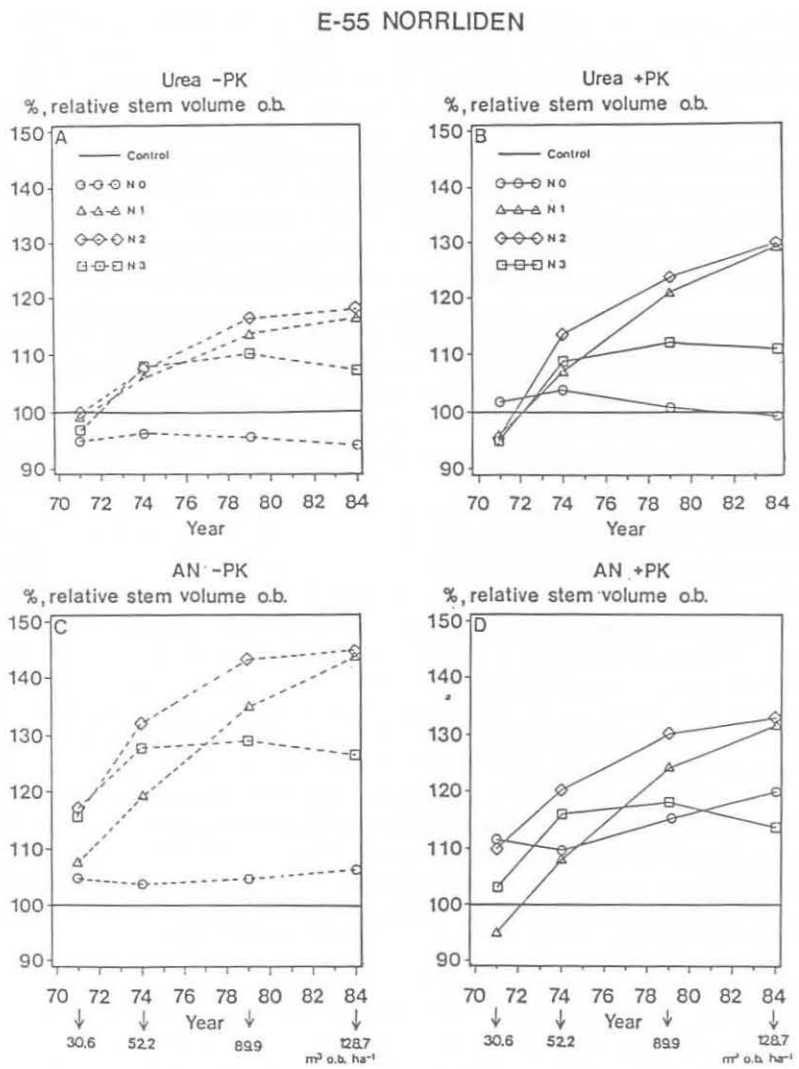

Fig. 4. Relative stem volumes over bark in different treatments of Experiments E55 Norrliden (Mean of six control plots = 100; values in cubic metres per hectare for these six plots given below lower diagrams). All values adjusted by analysis of covarance for initial differences in Bjørgung index (see Fig. 1). Symbols as in Fig. 3, except that broken lines mean without PK fertilizer, solid lines with PK.

sample trees and found rather similer slope changes in both types of diagrams.

The conclusions to be drawn at present from this experiments are the following:

1. Nitrogen is a limiting factor for forest growth on this site. All tested levels have increased growth, but there are clear indications that N3 (at present 90 kg N pr hectare and year) is above optimum, while there is little difference between N1 and N2 so far. The slope of the curves, however, suggests that also N2 may prove to be supraoptimum in a near future.
2. Although there have been significant interactions between nitrogen and PK in statistical tests of treatment effects on stem volume growth 1972–1979, positive with urea as source of nitrogen, negative with ammonium nitrate, it is difficult to decide from the present curves in Fig. 4 how important these effects are. The positive interaction urea/PK is easy to understand, and in a way expected.
3. Lesser vegetation has been changed by all nitrogen treatment, most drastically with N2 and N3 combined with PK.
4. Nitrogen fertilizers have increased mineral soil acidity. The effect is statistically significant at the levels N2 and N3, and may partly explain the negative effect of high nitrogen levels. The acidity changes have then to be taken in a wide sense, including decreases in the store of exchangeable base cations. In the humus layer, however, nitrogen fertilizers have decreased acidity in most cases. PK application has increased the base saturation, but with a negative interaction with nitrogen.
5. The results from this experiment, and others in the series «Optimum nutrient experiments», appear very relevant to the present situation in parts of Europe, with high deposition of nitrogen. The «critical loads» which can be deduced from Expt. E55 agree reasonably well with those deduced using quite different methods (Grennfelt & Nilsson 1988)
6. Although many of the a priori hypotheses behind the design of the experiment have proven correct (the dominating role of nitrogen in forest growth, the lesser importance of other nutrients, etc.), there have been some difficulties in interpretation of the whole causal chain of events, particularly because of the strong acidification of the mineral soil. These interpretation difficulties concern the answers to some of the basic ecosystem questions rather than the need for controlled experiments to simulate effects of high nitrogen deposition.

Summary

An experimental approach is essential for a good understanding of the ecosystem disturbances ascribed to air pollution. The interpretation of results from experimental as well as comparative studies is much easier, if a sufficiently detailed model for the impact on the ecosystem is used for the design of the investigation.

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